## **BLOOD LEUKOCYTES IN GM-CSF**

Culture 1d to adhere most monocytes Culture nonadherent cells 6d Feed with GM-CSF, culture 3d Rinse loose cells away

## AGGREGATES AFFIXED TO A DENSE ADHERENT MONOLAYER

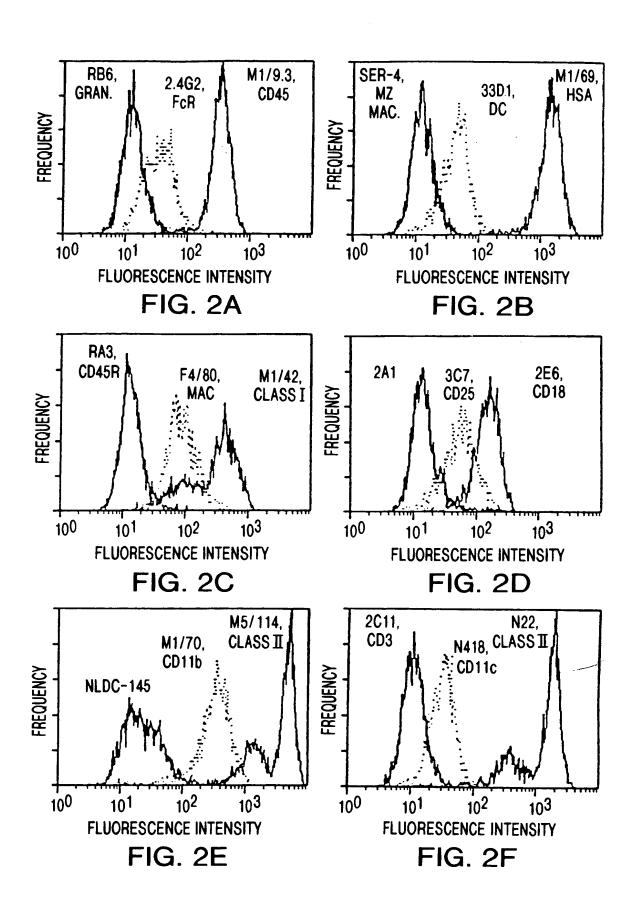
Dislodge aggregates with Pasteur pipettes Subculture one well to three in GM-CSF Culture 4-10d feeding q 3d DENDRITIC CELL AGGREGATES AFFIXED TO SCATTERED ADHERENT CELLS

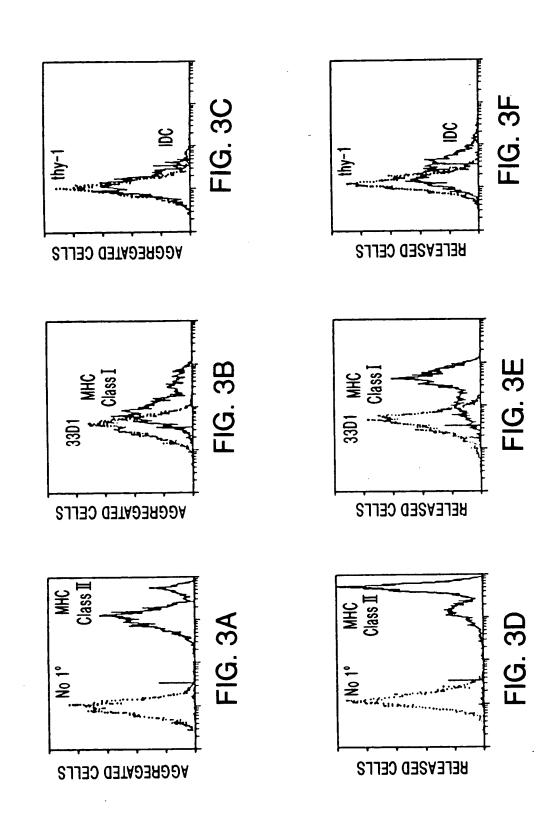
Subcultur

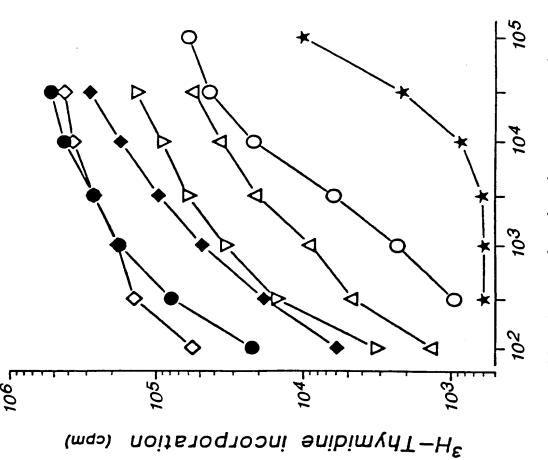
Subculture as above at 2-3 weeks

AGGREGATES ATTACHED TO A FEW ADHERENT CELLS PLUS RELEASED DENDRITIC CELLS

FIG. 1







Number of stimulator cells/well

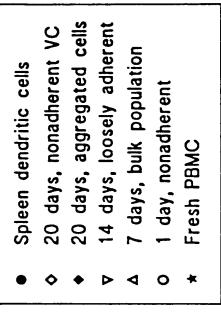
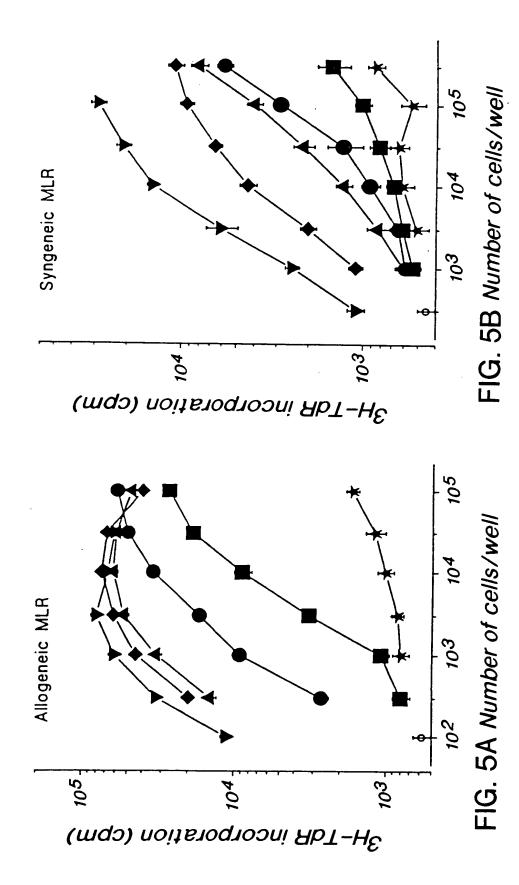
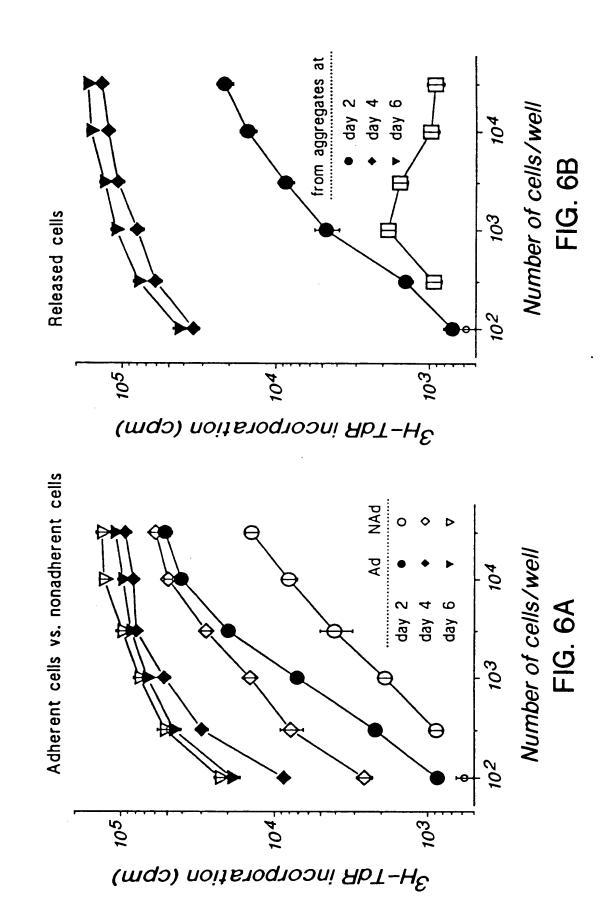
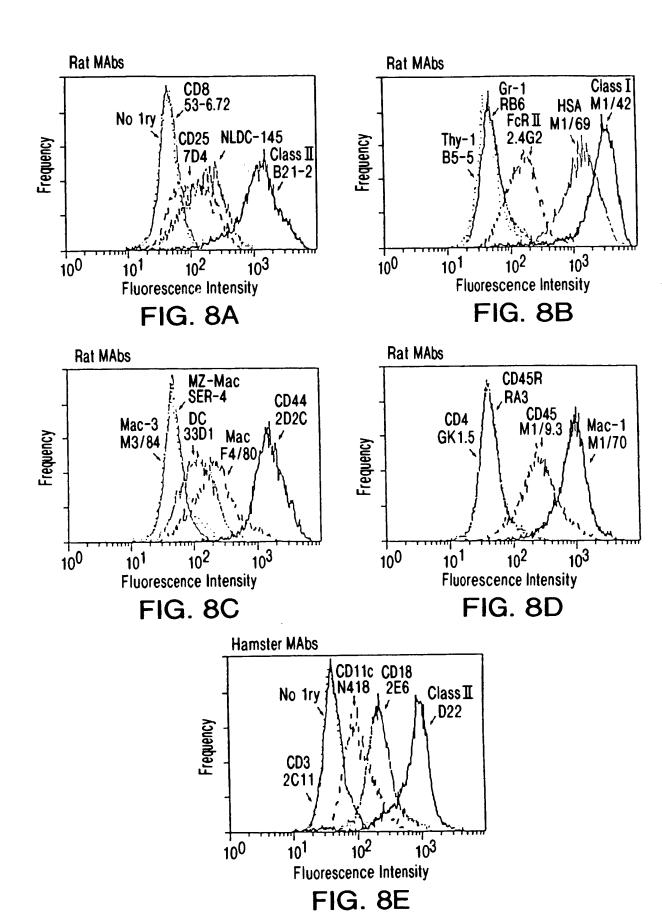


FIG. 4



\* 0 day, ■ 1 day, • 2 day, ▲ 4 day, ◆ 5 day, ▼ 6 day





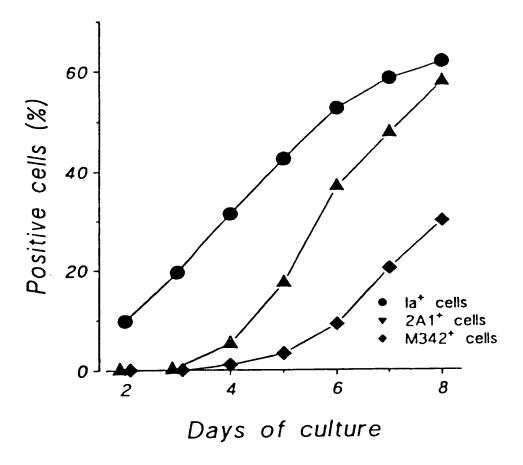
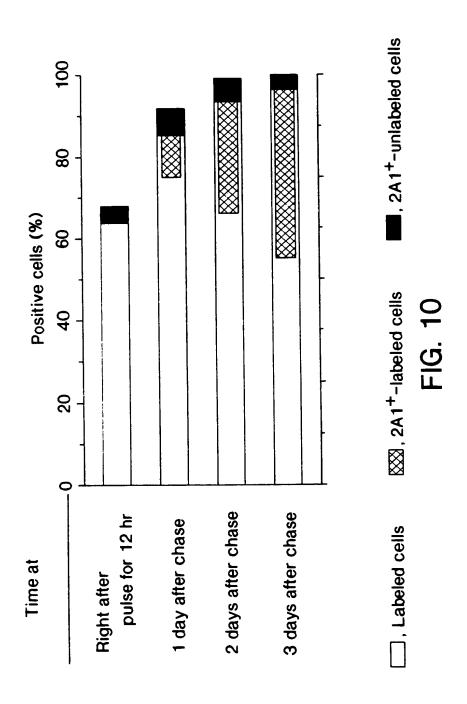


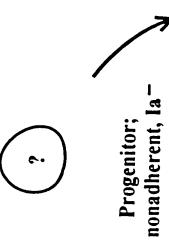
FIG. 9

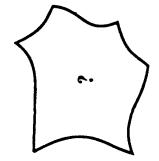


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Nonproliferating dendritic cells; nonadherent, Ia++

Proliferating
aggregate;
loosely adherent, la+
FIG. 11





Progenitor; adherent, ?la+

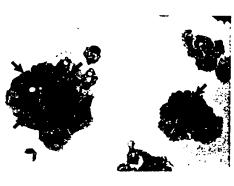
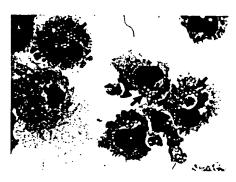


FIG.12A



F1G. 12B



FIG. 12C

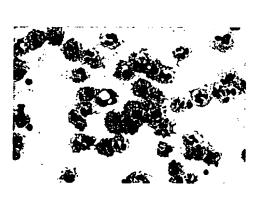


FIG. 12D



FIG.13A



FIG. 13B



FIG. 13C

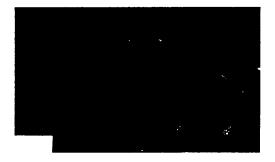


FIG. 13D

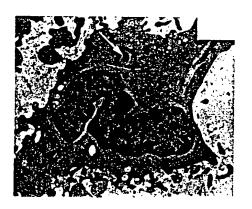


FIG.14A



FIG. 14B

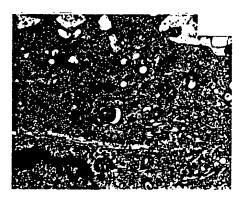


FIG. 14C

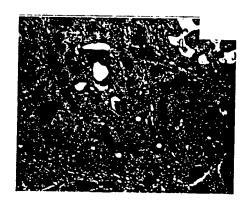
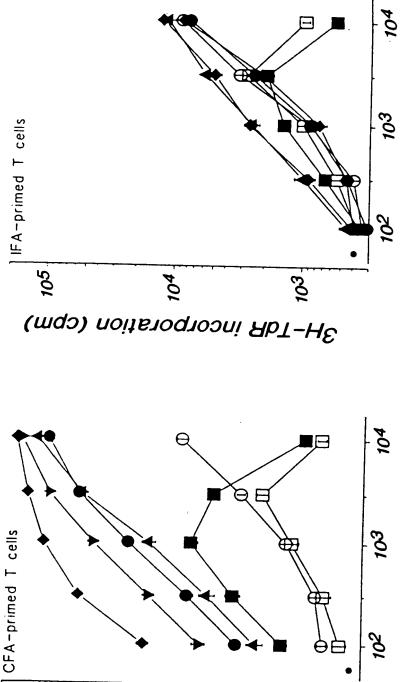


FIG. 14D



- Unpulsed, mature DC + PPD
  - Macrophage enriched
- Macrophage enriched + PPD
  - Immature, BCG-pulsed DC
    - Mature, BCG-pulsed DC
- Mature, BCG-pulsed and chased DC

105.1



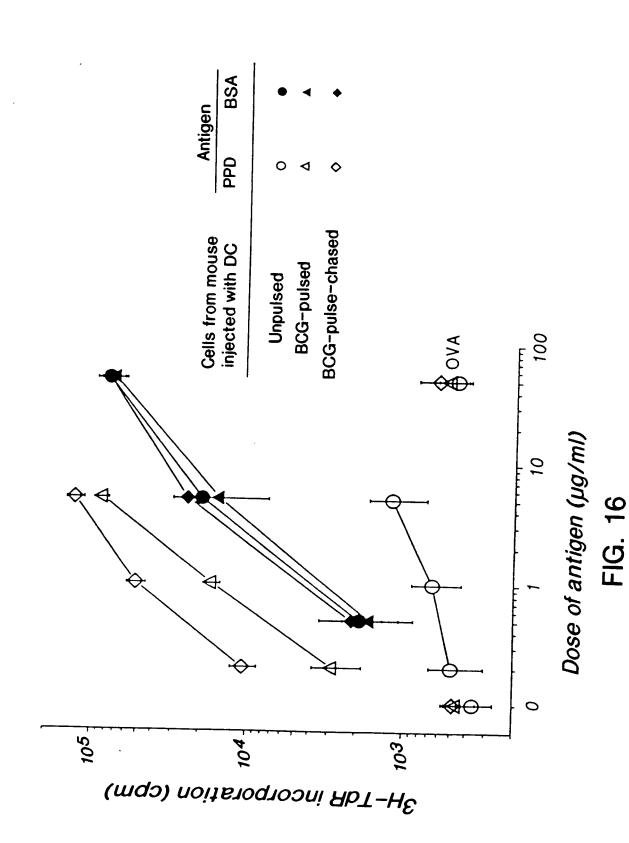
401

3H-TdR incorporation (cpm)

60

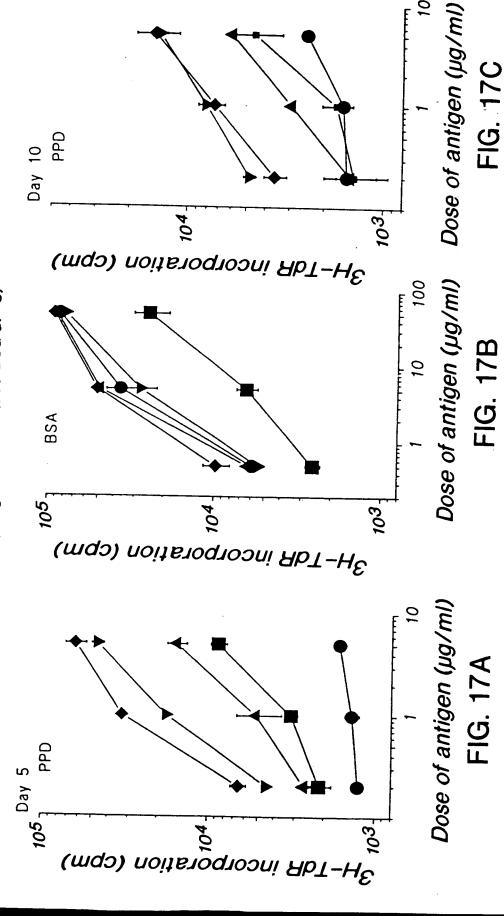
Number of dendritic cells/well FIG. 15A

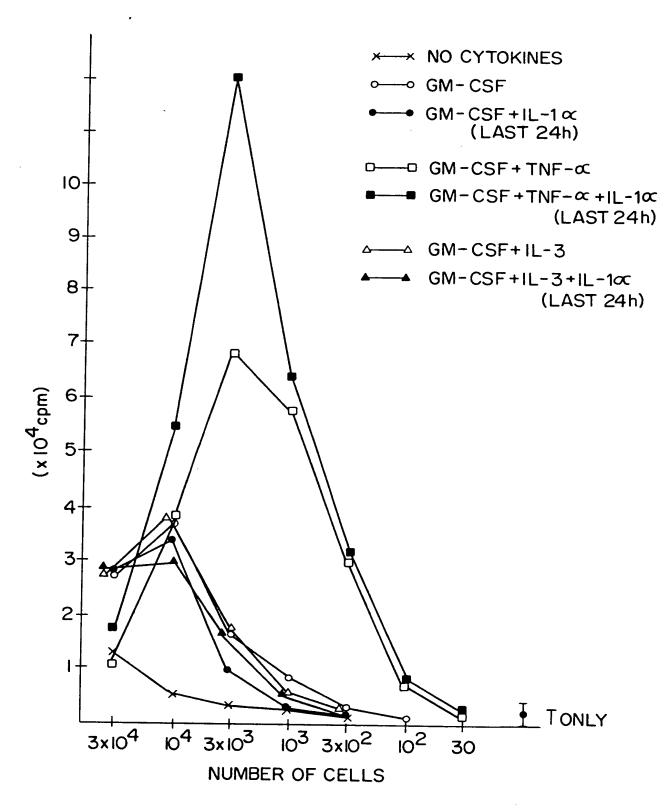
Number of dendritic cells/well FIG. 15B



Unpulsed mature DC

- Immature, BCG-pulsed (add BCG to growing d5-6 aggregates)
- Mature, BCG-pulsed and chased (add BCG d5-6, wash, chase to 8d) Macrophage-enriched (add BCG d7-8)





**FIG. 18A** 

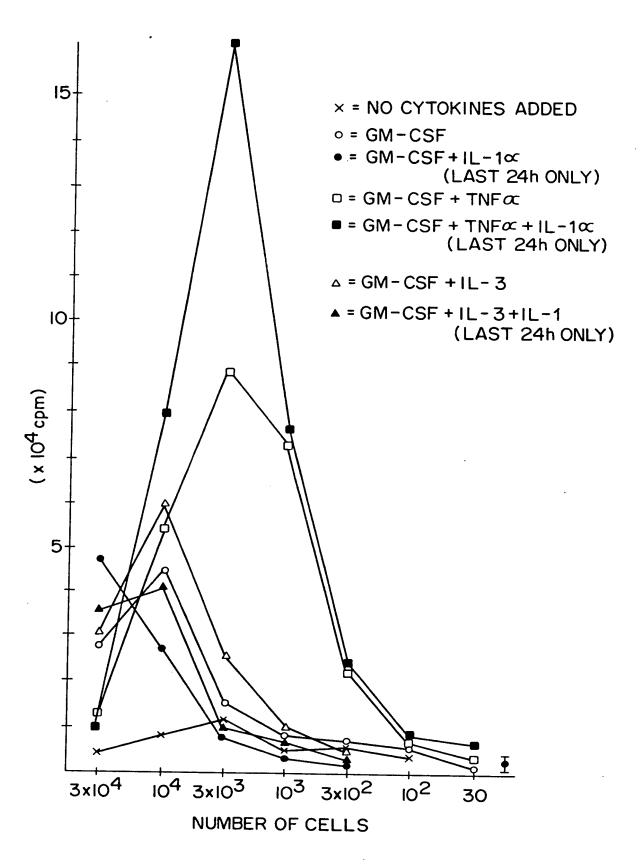


FIG. 18B

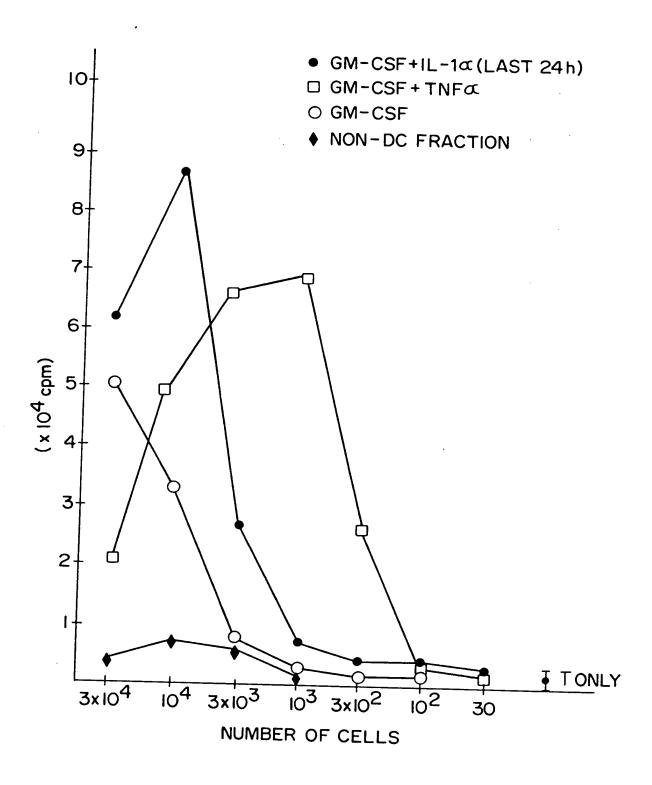


FIG. 18C